Sclerosant Foam Structure and Stability is Strongly Influenced by Liquid Air Fraction

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WHAT THIS PAPER ADDS

This study describes the influence of sclerosant concentration and liquid air fraction on foam stability, as well as on the size and count of bubbles following sclerosant foam injection. This will allow clinicians to make an informed decision about which concentration and liquid air fraction to use.

Objectives: To determine the effects of sclerosant foam preparation and composition on foam structure, the time course of liquid drainage, and foam coarsening.

Methods: Sodium tetradecyl sulphate (STS) and polidocanol (POL) foams were investigated in a range of concentrations (0.5—3%) and liquid-plus-air fractions (LAF; 1 + 2 to 1 + 8). Foam was injected into a vein simulation model (polyvinyl chloride tubing, inner diameter 3 mm, constant pressure 5—7 mmHg) filled with saline or blood. Liquid drainage, bubble count, and diameter were measured and documented by serial photography.

Results: Liquid drainage was faster in the vertical position than the horizontal one. In all variations, very small bubbles (diameter <30 μm) were generated initially that coarsened to form micro-foams (<250 μm). By 3 minutes mini-foams (>250 μm) and by 7.5 minutes macro-foams (>500 μm) were formed. Following injection, the upper regions of foam coarsened faster as liquid drained to the bottom of the vessel. Wet preparations produced significantly smaller bubbles. Low concentration POL foam produced significantly higher bubble counts and coarsened slower than STS.

Conclusions: Foam structure is strongly influenced by the LAF. Despite the initial formation of micro-bubbles in the syringe, mini- and macro-bubbles are formed in target vessels with time post-injection.

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INTRODUCTION

Detergent sclerosants sodium tetradecyl sulphate (STS) and polidocanol (POL) are surface active agents (surfactants) used in liquid or foam format to disrupt the endothelial lining of blood vessels with the ultimate aim of inducing endovascular occlusion.[1,2] Liquid sclerosants are heavily deactivated by blood components; hence, foam formats are preferred in the treatment of larger vessels owing to their increased potency.[3,4] The increased efficacy of foam is attributed to displacement of the intravascular blood and, hence, a greater exposure of the endothelial lining to the active agent.[3—5]

Aqueous foams are dispersions of gas in liquid, stabilised by surfactant adsorbed at the air—liquid interfaces.[6] Aqueous foams are out-of-equilibrium systems that evolve over time by gravitational drainage of liquid, coarsening, and film rupture. Foams coarsen because of pressure differences between bubbles of different sizes resulting in an increase of the mean bubble size. As aqueous foams coarsen at the top, a dry front propagates down through the foam and liquid emerges and accumulates at the bottom. Foam stability, liquid drainage, and foam coarsening are dependent on properties such as foam wetness, surfactant used, and bubble size.[6]

Sclerosant foams used in clinical practice are not standardised and are commonly generated using various techniques at the patient's bedside.[5,7] A dual syringe system is commonly used to generate foam by pumping a gas (such as room air) and the liquid detergent through a three-way stopcock. This study aimed to determine the effect of sclerosant foam preparation and composition on foam structure, and, in particular, the time course of liquid drainage and foam coarsening.

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MATERIALS AND METHODS

Definitions

Liquid drainage was defined as the percentage volume of liquid draining and separating from foam. Foam coarseness was defined using the diameter of bubbles as micro-foam (<250 μm), mini-foam (250–500 μm), and macro-foam (>500 μm).[2] Coarser foams were those containing larger bubble diameters. The liquid-plus-air fraction (LAF) investigated in this study included 1 + 2, 1 + 3, 1 + 4, and 1 + 8. Reference foam of 1 + 4 consisted of one part liquid and four parts air. "Wet" and "dry" were defined in comparative terms to 1 + 4. Hence, "wet" foams were those containing less air (1 + 2 and 1 + 3), while "dry" had a higher air component (1 + 8). A higher LAF implied a higher air component.

Materials

Materials included STS (FIBRO-VEIN 3% w/v; Australasian Medical and Scientific, NSW, Australia); POL (Aethoxysklerol 3% w/v, Chemische Fabrik Kreussler, Wiesbaden, Germany); sodium chloride 0.9% w/v (normal saline [NS]; Baxter Healthcare, NSW, Australia); 1-mL luer-lock syringe (Becton Dickinson, NJ, USA); 3-mL luer-slip syringe (Terumo, NJ, USA); three-way stopcock (Becton Dickinson); 5-μm Sterifix filter (B-Braun, Melsungen, Germany), 25-G (0.260 mm bore) needles (Becton Dickinson); 3-mm (inner diameter) polyvinyl chloride tubing (PVC) (Klein Infiltration Tube, HK Surgical, CA, USA); and a Nikon D70s digital camera with AF Micro-Nikkor 105-mm lens (Nikon, Tokyo, Japan).

Blood collection

Following informed consent, fresh donor whole blood was collected in citrate tubes (Vacutainer, 0.105 M; Becton Dickinson) from healthy volunteers taking no medications or supplements.

Sclerosant foam preparation

Liquid sclerosants were diluted with NS to achieve the desired concentrations. Foam was generated using a modified Tessari method.[7] Briefly, liquid sclerosant was drawn up in the 1-mL syringe and air in the 3-mL syringe to achieve the desired LAF. The two syringes were then assembled using a three-way stopcock with a filter placed on both syringes. Stopcock and filter assembly dead-space always consisted of air at the start of foam generation. The plungers were moved through 5–10 full strokes (see below) to disperse the air in liquid. A stroke was defined as a movement emptying and re-filling the syringe initially filled with liquid. The assembly was inverted once during foam preparation to optimise the mixing of air with the liquid sclerosant. The assembly of two filters and stopcock introduced a 0.35 ± 0.05 mL dead-space. Reported LAFs assume no dead-space.

Experimental set-up

All experiments and foam preparations were performed at ambient room temperature. Two sets of experiments were performed.

Injection syringe. Liquid drainage from sclerosant foam was measured in a 3-mL syringe to determine the effect of orientation, sclerosant type, and concentration. Foam preparations were allowed to rest in the syringe in either a vertical or horizontal position for 110 seconds. Horizontal syringes were then turned to the vertical position and fixed in place. This was to allow measuring the liquid volume using markers on the syringe. Serial photography was commenced for all syringes at 120 seconds until no further drainage could be detected. The percentage of liquid drained was calculated as the volume of the liquid layer at a given time divided by the total liquid volume.

Vein model. In a horizontal tube, images of foam were taken to assess flow behaviour and foam coarsening. The effects of sclerosant type, concentration, and LAF were determined. For these experiments, a vein model was constructed using a 110-cm PVC tube of 3 mm inner diameter, horizontal for 50 cm, with open elevated reservoirs positioned 30 cm from either edge. The tube was filled with saline or blood with an outlet back pressure calculated to be 5–7 mmHg (650–950 Pa). This pressure was selected to mimic saphenous vein pressure at the ankle level when supine.[4]

All foam combinations were injected 5–10 seconds after preparation into the vein model at mid-point. The needle tip was positioned just short of the base of the tube with the bevel facing upwards. Plunger displacement drove the foam into the tube (at consistent volumes indicated in the text) within 2 seconds at an angle of 25° (±5°) from the horizontal position. Between experiments the tube was cleared of residual bubbles with alternating water/air boluses.

Measurements

Overall foam behaviour. Overall foam behaviour and the direction of flow in the vein model were documented by serial photography.

Bubble count, diameter, and the rate of foam coarsening. Bubble count and bubble diameters were measured in the vein model at time points indicated in the text in three 8-mm-long regions in the upper half (1.5 mm high, i.e., 3 × 12 mm^2) of the foam as smaller bubbles in the lower half resolved poorly. Mean bubble counts were compared between sclerosants and either LAF or concentration.

Photography

Liquid drainage in the syringe and foam structure and behaviour in the vein model were documented by serial photography with a single pixel resolution of 0.015 mm (focus and zoom). A 50-mm-long region of the vein model

immediately anterior to the bevel was analysed. Imaging was obtained with a Nikon D70s digital single-lens reflex camera with AF Micro-Nikkor 105 mm lens fixed at 400 mm from the vein model.

Statistical analysis

Liquid drainage and largest consistent bubble diameter were assessed by multiple regression analysis using the R statistical computing package (v. 2.14.2; R-Project). Bubble counts were compared using Student *t* test with *a* significance threshold of $p < .05$, against a 3% concentration, 1 + 4 LAF, STS "reference" foam.

RESULTS

Liquid drainage

For all preparations, liquid drainage was initially decreased for horizontally orientated syringes compared with the vertical syringes (Fig. 1). However, after turning to the vertical position, within 180 seconds the liquid drainage in the horizontal syringes reached similar levels to the vertical ones.

Overall foam behaviour

Following injection into the vein model, foam flowed in both directions from the point of entry and occluded the full diameter of the vessel. Blood ingress commenced from the foam fronts and progressed below the foam/liquid layer resulting in the loss of the apparent initial occlusion. For the wettest foams (1 + 2), the full diameter of the vessel was not initially occluded and liquid drainage resulted in formation of a distinct lower liquid layer (Fig. 2).

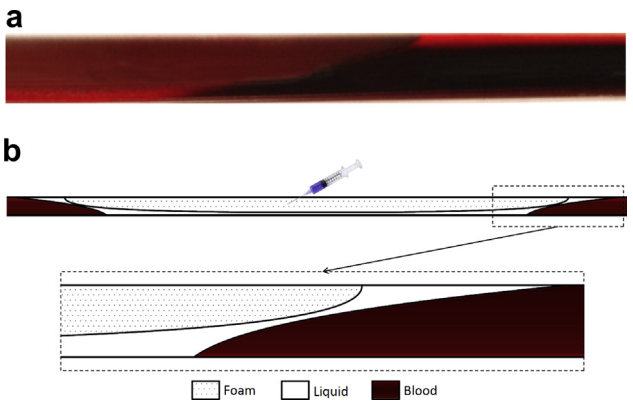

Figure 2. Blood ingress for wet (1 + 2) 3% foams. (a) The behaviour of 3% (1 + 2) sodium tetradecyl sulphate foam at the advancing edge, 2 minutes after injection of 1.5 mL of sclerosant foam. Liquid drainage from foam is apparent in lighter red regions. This drainage layer flowed below the bulk of the foam to form a lower liquid layer. Blood ingressed below the liquid layer at the foam front. (b) Schematic representation showing foam spreading 2 minutes post-injection; Inset: schematic representation showing foam (shaded), drained liquid (white), and blood (red).

Bubble count, diameter, and the rate of foam coarsening

In general, higher bubble counts corresponded with smaller bubble diameters. Immediately after the injection (Fig. 3), all foam preparations resulted in the formation of very small bubbles (≤30 μm) that coarsened to micro-bubbles (<250 μm). Upper regions of the foam coarsened more rapidly than lower regions. Mini-bubbles (250—500 μm) first became apparent in the upper regions by 3 minutes (Fig. 3) and macro-bubbles (>500 μm) by 7.5 minutes.

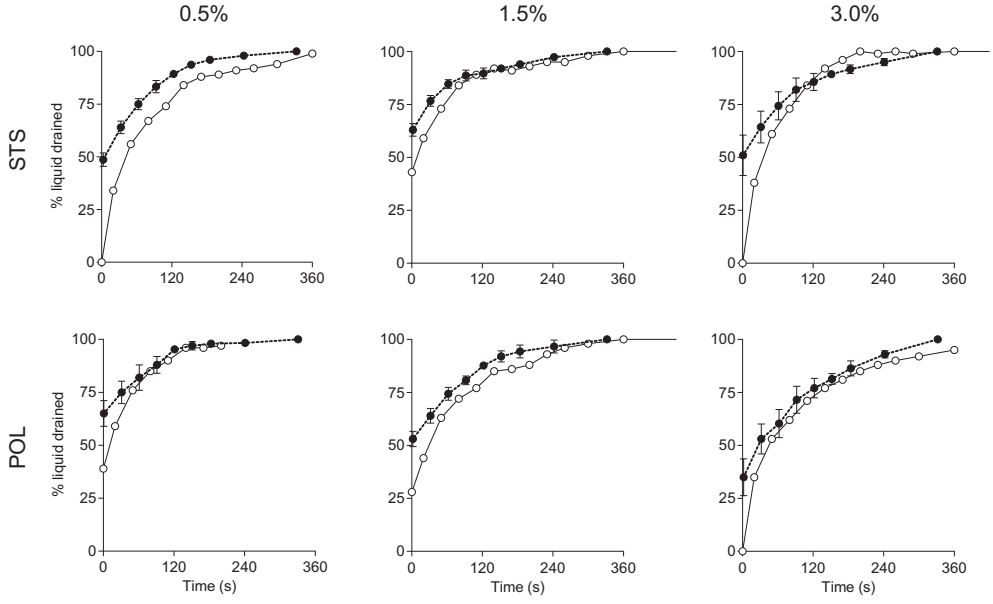

Figure 1. The effect of syringe orientation on liquid drainage. Liquid drainage from foam was assessed based on the orientation of the syringe following foam generation. Vertically-orientated syringes (●) were compared with horizontally-orientated syringes (○). The horizontal position was maintained for 110 seconds before the syringe was turned vertical for measurement. All measurements started after 120 seconds (shown here at time 0). *Note*. STS = sodium tetradecyl sulphate; POL = polidocanol.

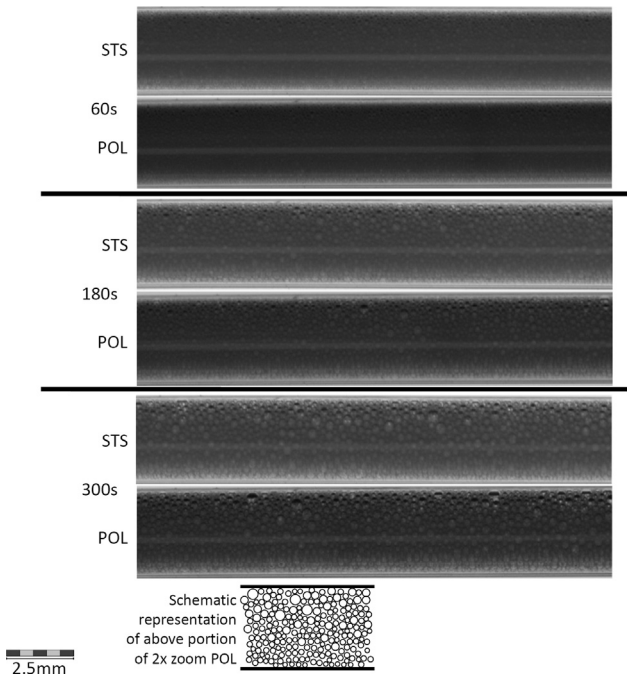

Figure 3. The time course of foam coarsening. Foam coarsening with time is shown after injection of 1.5 mL of 3% (1 + 4) sodium tetradecyl sulphate (STS) or polidocanol (POL). Mini-bubbles (250–500 μm) were apparent 3 minutes after injection, with a wide distribution of bubble diameters. No differences between the sclerosants are apparent. Larger bubbles focus at the top of the tube over time. Foam appears dark as bubbles reflect dark walls of experimental rig.

When injected into a saline model, low-concentration STS foams coarsened faster than high concentration STS foams, whereas, for POL, high-concentration foams coarsened fastest (Fig. 4). When injected into blood, there was no significant difference between bubble diameters for 0.5%, 1.5%, and 3% STS; however, POL at 0.5% produced significantly smaller bubbles than 1.5% and 3% POL (Fig. 5a). The decreased bubble diameter seen at 0.5% POL corresponded to a significantly increased bubble count (Fig. 5b).

The wettest preparations had a smaller bubble diameter and coarsened more slowly, while the dry foams had lower mean bubble counts and larger bubble diameter in all cases (Fig. 5c, d). POL produced significantly higher bubble counts than STS for the wettest foams (1 + 3).

DISCUSSION

In this study, we investigated the effect of sclerosant foam preparation and composition on foam structure, the time course of liquid drainage, and foam coarsening in a vein simulation model constructed out of PVC tubing. Here we report the significant effect of foam wetness on liquid drainage, the time course of foam coarsening, and bubble diameter.

We assessed the effect of foam composition on liquid drainage and showed that liquid drainage occurred more slowly in horizontal syringes than in the vertical ones. The delay in drainage for syringes stored horizontally is consistent with previous foam drainage studies that predict taller foams draining faster owing to the reduced capillary effects.[8,9] Drainage increases the coarsening rate and results in increasingly dry foam with height.[6,8,9] Therefore, once foam is generated, syringes should be kept in the horizontal position to reduce liquid drainage and foam coarsening.

Following the injection into the vein model, all preparations but the wettest foams (1 + 2) appeared to occlude the full diameter of the vessel initially. The wettest foams demonstrated the smallest bubble diameters; hence, this observation may indicate a trade-off between keeping bubbles small while maintaining a stable occlusion of the vessel with foam. In time, three layers of coarsened foam, liquid, and blood were observed in the vein model with blood ingress flowing from each foam front below the drained liquid layer. Equal blood ingress from both sides is not unexpected as there was no pressure gradient across the vessel. The separation into three layers reflects the density of each layer, with air being the least dense and foam staying at the top, and blood being the densest layer, ingressing below the foam and liquid sclerosant layers. In a recent study, we determined the density of the liquid and foam sclerosants, and demonstrated a lower density for foam sclerosants when compared with liquid agents and blood.[10]

In this study, all preparations, irrespective of LAF or the type or concentration of sclerosants foam, produced very small bubbles (<30 μm) that coarsened to form micro-foams (<250 μm). Mini-foams (250–500 μm) were formed by 3 minutes and by 7.5 minutes macro-foams (>500 μm) were formed. In general, smaller bubbles were generated by wetter preparations, and smaller bubble size correlated with a higher bubble count. Drier foams coarsened faster resulting in a faster formation of macro-bubbles. Bubble size increased with height in the coarsening foams to produce a size gradient, as a result of drainage, with larger bubbles coarsening at the top and smaller bubbles persisting at the bottom of the vessel.

The coarsening rate was initially faster for STS than POL, but both agents arrived at similar means at 3 minutes. Concentration influenced coarsening rates differently for each sclerosant. The coarsening rate was slower at lower POL but higher STS concentrations. Overall, the effect of concentration was small, suggesting the 0.5–3% range is well above the critical micellar concentration—the concentration above which surface tension is approximately constant. Bubble diameter was smaller for wetter foams initially and over time.

This study may have a number of practical implications. Higher liquid content resulted in smaller bubbles and allowed bubbles within the foam to move more easily. Hence, LAFs of 1 + 3 and 1 + 4 are best optimised to occlude larger vessels, while minimising bubble size. Clinically, micro-bubbles are preferred to macro-bubbles, as micro-foams increase the exposure of the endothelial lining to the active agent.[5] Furthermore, macro-bubbles are more likely to occlude small cerebral arteries than micro-bubbles,

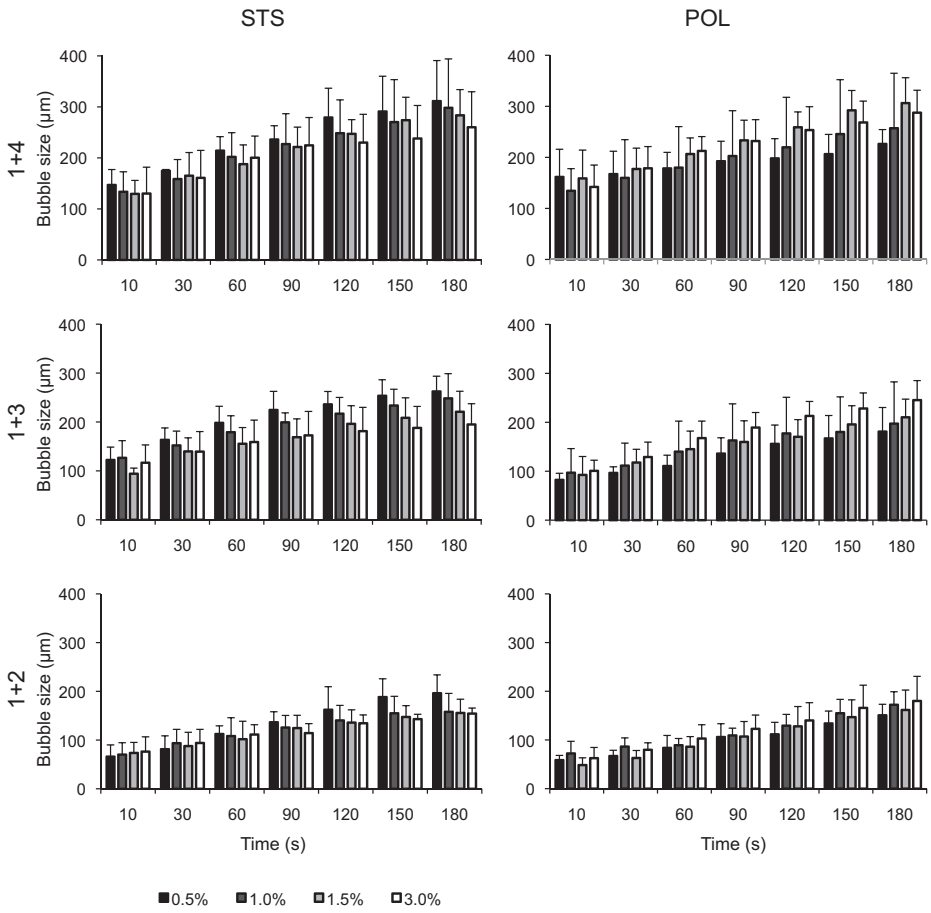

Figure 4. The effect of liquid air fraction and sclerosant concentration on foam coarsening. The effect of liquid air fraction and sclerosant concentration on bubble diameter was assessed for 3 minutes post injection of 1 mL of sodium tetradecyl sulphate (STS) and polidocanol (POL) into the vein simulation model containing normal saline ($n \geq 3$).

resulting in gas embolism. Hence, the ideal sclerosant foam should generate small bubbles that coarsen slowly and absorb rapidly. Given the present findings, sclerosant foams should be injected within 3 minutes of preparation to avoid larger bubbles. Using micro-bubble preparations, however, is not a safeguard against intravascular coarsening of the injected foam. The clinical intravascular evolution of sclerosant foams and the ultimate fate of the injected bubbles may not parallel these in vitro findings and should be investigated in future clinical studies.

The choice of sclerosant and concentration influences the bubble shape and size. In this study, high concentration (3%) STS and low concentration (0.5%) POL produced smaller bubbles that coarsened slowly, whereas low-concentration (0.5%) STS and high-concentration (3%) POL produced larger bubbles that coarsened rapidly. The bubble size and shape (polyhedral vs. spherical), and the three-dimensional structure of the aqueous foam varies according to the sclerosant type and characteristics such as charge and viscosity. In a recent study we have studied the relationship between viscosity and temperature variations to foam structure and stability.[10,11] Further studies are underway to investigate the bubble shape and micro-structure of sclerosant foams.

The ingress of blood from the foam fronts and formation of a mixed liquid blood layer would result in mixing, dilution, and deactivation of the sclerosant and a drop in the final concentration.[3] This would have significant clinical relevance as lower concentrations are associated with a procoagulant profile,[12] which has been clinically detectable at the foam front.[13] Therefore, wetter foams are least suitable in achieving a stable occlusion in larger vessels. It would be clinically prudent to reduce the diameter of larger vessels before the administration of foam sclerosants. This can be achieved by using methods such as infiltration of tumescent fluid in the perivascular space before the administration of the sclerosing foam. Induction of adequate venous spasm would, in particular, help the wetter foams to be more occlusive and reduce the mixing and deactivation at the foam front.

This study had several limitations. We investigated room air as the foaming gas and used 1-mL/3-mL syringe combinations to generate the foam. Other gases, such as carbon dioxide (CO_2) or a combination of CO_2/oxygen, as well as other syringe combinations (e.g., 3-mL/5-mL; 5-mL/10-mL), are also used in the clinical practice and should be investigated in future studies. A combination of higher resolution imaging and image analysis techniques would be required

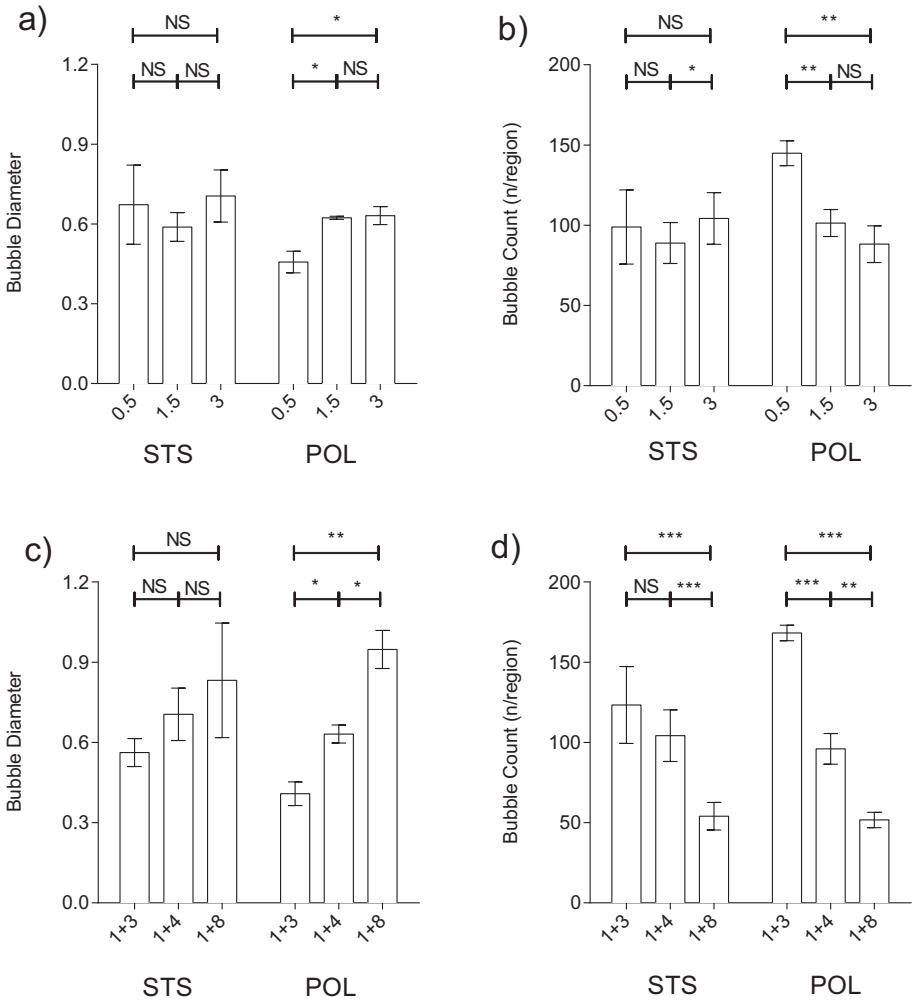

Figure 5. Bubble count and diameter following foam injection. The effect of (a, b) sclerosant concentration and liquid air fraction (LAF) (c, d) on mean bubble diameters (a, c) and bubble count (b, d) at 7.5 minutes for sodium tetradecyl sulphate (STS) and polidocanol (POL) foams injected into a vein model filled with blood. The effect of sclerosant concentration was tested on LAF 1 + 4, and the effects of LAF was tested at 3% sclerosant concentration. Note. *$p < .05$, **$p < .01$, ***$p < .001$; $n \geq 3$. NS = not significant.

to achieve a better understanding of bubble size. The vein model used in this study had a fixed diameter whereas real vessels change size in response to temperature variations, environmental factors, or vasoactive mediators and stimuli.

In conclusion, while it is known that injected foam is composed of micro-bubbles, if this foam remains cohesive for minutes then significantly larger bubbles form. Foam structure is strongly influenced by LAF. Higher liquid content results in formation of micro-bubbles that coarsen slowly, while higher gas content results in formation of dry foams that coarsen more rapidly. The ingress of blood at the foam front and liquid drainage from foam results in formation of distinct layers of foam, liquid, and blood in the vessel.

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CONFLICT OF INTEREST

None.

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